THE ROLE OF OXYSTEROLS IN A COMPUTATIONAL STEROIDOGENESIS MODEL OF HUMAN H295R CELLS TO IMPROVE PREDICTABILITY OF BIOCHEMICAL RESPONSES TO ENDOCRINE DISTUPTORS

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Steroids, which have an important role in a wide range of physiological processes, are synthesized primarily in the gonads and adrenal glands through a series of enzyme-mediated reactions. The activity of steroidogenic enzymes can be altered by a variety of endocrine disruptors (ED), some of which are environmental contaminants. We are developing a dynamic computational model of the metabolic network of adrenal steroidogenesis in a human H295R cell line to predict the synthesis and secretion of adrenocortical steroids (e.g. mineralocorticoids, glucocorticoids, androgens and estrogens), and the biochemical response to ED. We previously developed a deterministic model which describes the biosynthetic pathways for the conversion of cholesterol to adrenocortical steroids, and the kinetics for enzyme inhibition by the ED, metyrapone (MET). In this study, we extended the model by adding the pathway of oxysterol biosynthesis. Oxysterols are endogenous products of cholesterol unrelated to steroidogenesis. Experiments were performed to measure concentrations of cholesterol and 14 steroid concentrations in human H295R cells using LC/MS/MS and ELISA methods. Model parameters were estimated using an iterative optimization algorithm. Results show that the model fit improved with the extended model. Model predictions closely correspond to time-course measurements of both cholesterol and steroid concentrations from control and dose-response experiments with MET. Our study demonstrates the feasibility of using the computational model of adrenal steroidogenesis to predict the *in vitro* adrenocortical steroid concentrations from human H295R cells. This capability could be useful to help define mechanisms of action for poorly characterized chemicals and mixtures in support of the H295R steroidogenesis screening system, and predictive risk assessments. *Although this work was reviewed by EPA and approved for publication, it may not necessarily reflect official Agency policy.* (M. Breen was supported by the NCSU/EPA Cooperative Training Program in Environmental Sciences Research, Training Agreement CT833235-01-0 with North Carolina State University.)